EVALUATION OF SELENIUM'S ROLE IN HEAVY METAL BIOACCUMULATION AND TOXICITY

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Project Description

Environmental mercury can either bioaccumulate within an aquatic food chain as the readily absorbed and highly toxic form methylmercury (MeHg) or biologically retire as insoluble and inert forms such as mercury sulfide (HgS) and mercury selenide (HgSe). Based on ongoing CATM research, it appears that the extraordinarily high binding affinity between Se and Hg is involved in many important aspects of MeHg bioaccumulation and pathophysiology. This project examines these interactions from multiple perspectives in order to assess the environmental benefits of Se binding with Hg and any physiological harm that can occur as a result of Hg binding with Se.

Goal

In order to better understand how Se limits Hg accumulation and how Hg harms Se physiology, an integrated research program was needed. Our prior research utilized animal models to study Se-dependent protection against Hg toxicity and invertebrates to assess Se-dependent Hg retirement from aquatic ecosystems. To continue with an integrated approach to analysis, the goals of this project are threefold: to assess environmental factors of Hg and Se accumulation by analyzing plants and invertebrates and the potential ways of remotely detecting Hg and Se accumulation in plants, to investigate the influence of Se status on MeHg toxicity at the cellular level in time-dose dependent studies, and to investigate human effects of Hg accumulation through the examination of heart tissues of human patients with dilated cardiomyopathy.

Rationale

The importance of the Se–Hg interaction has many environmental and physiological implications that can influence MeHg exposure risks. Selenium, an important nutrient present in many foods including fish, is known to counteract Hg toxicity. It had previously been thought that selenium's potent protective effect against Hg toxicity was due to its exceptional binding affinity for Hg. However, a different perspective on this interaction suggests MeHg toxicity is actually the result of this binding affinity. This mechanism involves mercury sequestering intracellular selenium, thereby inhibiting the formation of vital selenoenzymes in the central nervous system. If this is the case, the greatest risk from MeHg exposure

will occur in Se-poor populations whose diets originate from locally grown foods and freshwater fish. The Se-rich fields and pastures of the midsection of North America produce Se-rich foods that are broadly distributed through the centralized food distribution system. However, populations and individuals living in Se-poor regions may be at risk of having lower Se status if they primarily consume locally grown foods. Selenium-poor regions of the world include much of Northern Europe, most of Africa, and many parts of Asia. Because of the particularly low soil Se levels in Finland and New Zealand, risks associated with Hg exposure there would be far more pronounced than elsewhere. Populations in these countries and other Se-poor regions need to be cautious of consuming diets exclusively comprising foods grown in their local areas, since a low Se status will offer less Se-dependent protection against MeHg exposure. Fish originating from lakes in regions with low Se availability will be particularly risky to consume since these lakes will tend to display increased MeHg bioaccumulation in fish, and there will be far less Se available to counteract the toxicity of the MeHg that is consumed.

Selenium availability for uptake by plant life in the aquatic environment is likely to have an important effect on Hg bioaccumulation in the food chain. Formation of HgSe is minimal in the water column but greatly potentiated by the molecular species of Hg and Se that occur within living cells. Plant life, especially in the cases of Hg hyperaccumulator plant species, are likely to contribute to Hg retirement through HgSe formation as a result of biochemical processes that parallel those occurring in animal cells. Invertebrates that form the base of the aquatic food chain will contribute to Hg retirement also by facilitating HgSe formation. Likewise, selenium availability and uptake by plant and animal life in the aquatic environment will diminish the bioaccumulation of Hg in the food chain. Since HgSe is insoluble and extremely resistant to dissociation, Hg occurring in this form in the tissues of prey animals is poorly absorbed by predators that consume them. Therefore, if MeHg accumulation is reduced in the insects that make up 90% of the food consumed by fish, it will diminish the amount of MeHg accumulation in fish.

Currently, there are no techniques available that allow a rapid, precise, and inexpensive means of identifying areas that have high concentrations of heavy metals. Extensive sampling is currently needed to map soil Se and Hg concentrations. However, spectroscopic analysis of vegetation may provide a means of detection that can be used to identify either isolated or regional distributions of these elements. Remote sensing has shown that Se-rich soybeans have a spectral reflectance shift to shorter wavelengths in the long-wavelength edge of the chlorophyll absorption band centered at 680 nm and higher reflectance in the 550–650-nm region. Therefore, Se incorporation in plants causes a specific shift in reflectance of light at 660 nm. This spectral shift may enable rapid and remote detection of Se distribution, thereby establishing a means of identifying areas of high vs. low Se soil availability. This technology is currently being assessed in a related CATM project, and if sufficiently sensitive, sensing through satellite imaging may eventually become possible.

Studies performed on men in eastern Finland indicate Hg exposure from consistently eating local lake fish is associated with increased cardiovascular risk. Since eastern Finland has very poor Se availability, low Se and high MeHg contents in fish from that region may have caused impairments in selenoenzyme functions that may have contributed to cardiovascular impairments as a result of diminished free radical detoxification. The influence of Se on Hg methylation/demethylation rates and Hg-dependent losses of available Se for sustaining selenoenzyme activities are important metabolic interactions that need to be assessed for understanding these pathologies. However, these molecular effects are difficult to study in human or animal models. Therefore, for a thorough investigation of mechanisms involved in any biological issue, cell culture must be included. The specific advantage of cell culture methods is that they reduce the physiological and biochemical constraints imposed by in vivo studies. Cell culture offers a completely controlled environment, thereby providing an opportunity to investigate very specific research questions and biological mechanisms.

The toxic effects of MeHg and inorganic Hg in cultured cells vary somewhat with cell type and occur at slightly different concentrations (1), but differences in the molecular mechanisms associated with their respective pathologies have not been adequately studied. The entry of inorganic mercury into the brain is largely prevented by the blood–brain barrier, whereas organic mercury can pass the barrier more easily (2). However, it is interesting to note that a significant fraction of the total Hg bound in animal and human brains (3–5) is inorganic, a portion of which appears to originate from MeHg demethylation (6). In the brain, glial cells appear to be responsible for this demethylation activity (7). Methylmercury at a concentration of 0.1 μM has shown to be lethal in cell cultures (8), but no one has measured the direct effects of MeHg exposure on Se-dependent enzyme activities in cultured cells.

Curiously, unusually high concentrations of Hg and antimony (Sb) accumulation were reported in heart tissues of human patients suffering from dilated cardiomyopathy (DCM). These levels have not been confirmed and may be due to analytical errors resulting from the small sample sizes obtained using biopsy methods. However, if validated, this phenomenon could lead to further directions in understanding MeHg-associated cardiovascular disease. Since almost half of all heart transplants are performed on DCM patients, a significant potential source of heart tissue for study would be the explanted heart removed during transplantation. Larger tissue samples could provide an opportunity for the highest-quality analytical evaluations so that these levels can be reassessed and validated. Likewise, selenium levels will also be assessed, since selenium has an important role in Hg toxicity and Hg accumulation seldom occurs without formation of HgSe.

Approach

Mercury–Selenium Interactions in Plants and Invertebrates

Formation of HgSe is being investigated in plants known to hyperaccumulate Hg and Se and in a model invertebrate species (Achaeta domesticus; house crickets). Since it enters the selenium cycle directly without requiring initial degradation, sodium selenite is the most commonly used form in Se nutrition studies. Therefore, although it is not a normal form found in food, it directly reflects the effects of the forms of selenium naturally present in foods. Food forms are a mixture of organic forms including selenomethionine and selenocysteine. Therefore, these studies compared the effectiveness of organic and inorganic forms of selenium in protecting against MeHg toxicity. The crickets from the Trophic Level 1 study were fed torula yeast-based diets prepared with graduated concentrations (0.1, 1.0, or 10 μmol Se/kg) of inorganic selenium. The selenium absorbed and retained in the tissues of these crickets is almost exclusively present as selenocysteine since animal tissues are unable to form selenomethionine. Absorption and retention of Se was assessed in the presence and absence of 0.5 μmol MeHg/kg added to the diets. By the end of the Trophic Level 1 phase of the study, Se-dependent effects on Hg retention was directly assessed in freeze-dried cricket tissues. Any MeHg-dependent effects on Se retention was determined by comparing Se present in tissues of the 1-μmol Se, 0.5-μmol MeHg/kg test group to the amount of Se present in tissues of the control group fed 1 μmol Se/kg without any added MeHg.

Since crickets are opportunistic cannibals, crickets in the Trophic Level 2 study are currently being fed selenocysteine in the form of ground crickets from Trophic Level 1. To ensure their diets are nutritionally complete, their remains are mixed with Se-free torula yeast-based diets to ensure adequate intakes of necessary nutrients.

To investigate the spectral reflectance shift of vegetation induced by Se (and Hg binding of Se), rabbit foot grass and water hyacinth are being grown under controlled conditions using hydroponic solutions. The plants are provided basal nutrient solutions prepared with or without Hg and Se added at

graduated concentrations. As the plants grow, Hg- and Se-dependent spectral reflectance of leaves is being measured by spectrophotometry.

MeHg–Se Interactions at the Cellular Level

To understand the environmental and epidemiological effects of mercury exposure, it is important to understand mercury mechanisms at the cellular level, particularly in the central nervous system. The approach for this study was to use cell cultures grown in the presence of media containing graduated concentrations of Se and subject them to MeHg and inorganic Hg added at incremental log concentrations ranging from unexposed to toxic levels. Parallel studies of these Se-conditioned and Hg-exposed cultures are being challenged for specified time periods to establish the time- and dose-dependent effects of MeHg toxicity and Se's protective effects against toxicity. The end points measured include cell proliferation rates, viability, mercury and selenium concentrations, Se-dependent enzyme activities, and rates of MeHg demethylation.

Mercury's Effects on Cardiovascular Disease

Working with research pathologists and surgeons at the Mayo Clinic in Rochester, Minnesota, tissue samples have been collected from the explanted hearts removed from patients undergoing heart transplantation in response to ischemic heart disease (IHD) and DCM. The Hg and Se contents of these samples are being assessed in comparison to heart tissues without recognizable pathology collected at autopsy from patients dying of accidental causes. The elemental analysis data are being compiled and statistically compared in order to confirm or refute the reported observations of unusually high Hg levels in DCM hearts.

Progress/Status

Mercury–Selenium Interactions in Invertebrates

The crickets from the first food chain study examined the interactive effects of mercury and selenium on one another's absorption. The biomass of these crickets was retained in dry form, mixed with Se-free torula yeast, and fed to the next trophic level of crickets.

Results are presented in Figures 1a and 1b. The figures show that crickets that were fed crickets grown on mercury-free diets grew better ($p < 0.05$) than crickets fed powdered diets composed of crickets grown on 0.5 µmol Hg/0.01 µmol Se or 0.5 µmol Hg/1.0 µmol Se, but equivalent to crickets fed crickets grown on 0.5 µmol Hg/10 µmol Se. The amounts of Hg bioaccumulated in crickets in Food Chain II were not statistically different, but their Se contents were proportional to the dietary Se levels fed crickets in Food Chain I ($F = 4.77$, $p < 0.03$). The crickets from Food Chain II will be used as fish food in the next phase of this study.

Mercury–Selenium Interactions in Plants

The best analytical precision in trace element analysis is attained using dry sampling methods, thus avoiding potential variance due to differences in water contents of individual samples. Previous work had established Hg and Se concentrations in plant materials were not lost during heat-drying since observed concentrations were equivalent if freeze-drying or wet-sampling was performed. Two different plant species have been tested, *Polypogon monspeliensis* and *Eichhornia crassipes*. Leaves and stems of *P. monspeliensis* collected at the end of Task 1 were dried for 24 hours at 70°C in a laboratory oven prior to

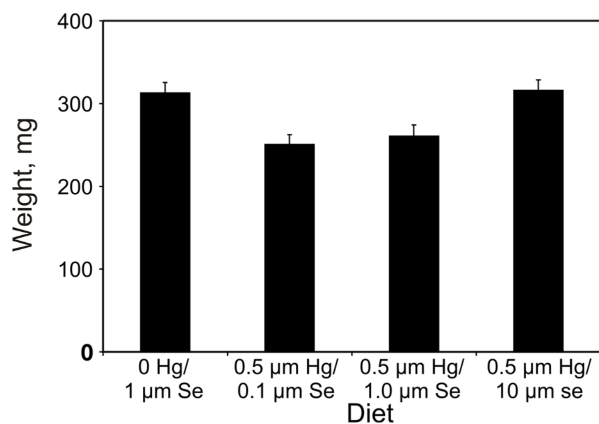

Figure 1a. Cricket body weights.

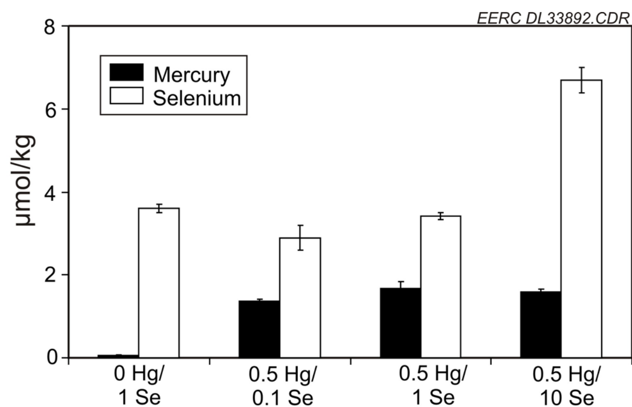

Figure 1b. Mercury and selenium in crickets.

Figure 1. Impact of Hg/Se on cricket growth

sampling. Because of their higher water contents and denser tissues, leaves, stems, and roots of *E. crassipes* collected at the end of Task 2 were oven dried for 48 hours prior to sampling for elemental analysis.

Germination of *Polypogon monspeliensis* seeds obtained from the National Plant Germplasm Resources Laboratory was less than 20%, so *P. monspeliensis* seeds were instead obtained from Sandeman Seeds, in Lalongue, France. The germination rate of these seeds (sets of 100 seeds examined in triplicate) was 95 ± 2%. Twenty milligrams of P. monspeliensis seeds was planted in potting soil in 12-inch-diameter pots. Plants were watered with 200 mL of DynaGro daily throughout the study. Pots containing seeded soil were covered with transparent lids until germination was complete. Plants were grown indoors in ambient sunlight under temperature-controlled conditions (20°–25°C) during the entire study.

When the plants were approximately 4 inches high, the supplementation phase of the study began, using a fully crossed 3 × 3 factorial (nine growth treatment groups) study design to examine dose effects of Se and Hg on one another's elemental uptake. During this phase, in addition to the 200 mL of diluted Dyna-Gro solutions, plants were supplemented with weekly additions of solutions prepared with 100 mL of graduated concentrations of mercury and selenium (50 mL of supplementation solution of each element). The solutions contained low, medium, or high levels of Se as sodium selenite (0.01, 0.1, and 1.0 mmol Na_2SeO_4/kg, respectively), and low, medium, or high levels of Hg as mercury chloride (0.01, 0.1, and 1.0 mmol $HgCl_2$/kg, respectively). Plants were supplemented weekly for ~8 weeks. At the end of the study, plant leaves and stems were harvested from each treatment group and stored in trace metal-free plastic bags at -85°C until ready for analysis.

The amount of Se in leaves of P. monspeliensis plants grown without appreciable amounts of added Se or Hg was 0.83 ± 0.24 µmol Se/kg (0.070 ± 0.02 µg Se/g). As seen in Panel A of Figure 2, addition of 100- or 300-mM Se solutions resulted in minor increases in Se incorporation, but when Hg was added, Se accumulation in leaves was greatly increased. The roots on these plants have not been analyzed yet, so it is not known whether the effect of Hg on Se levels in the plants represents an accentuated uptake from the soil or redistribution from the roots.

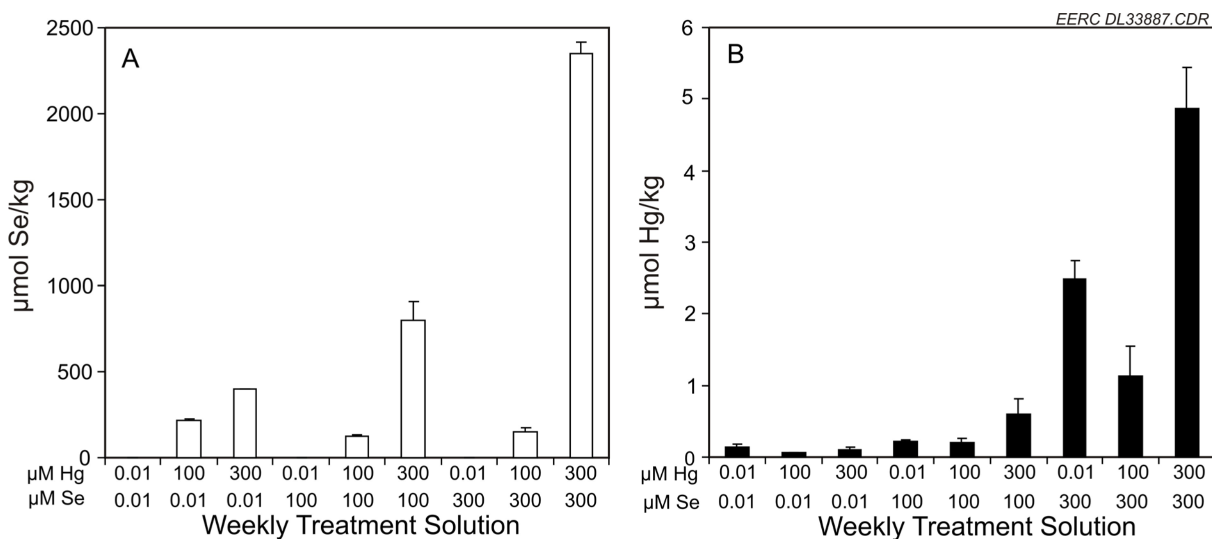

Figure 2. Mercury and selenium in P. monspeliensis grown in soil treated weekly with solutions containing the indicated concentrations of Hg and Se.

The amount of Hg in leaves of *P. monspeliensis* plants grown without appreciable amounts of added Se or Hg was 0.15 ± 0.04 µmol Se/kg (0.029 ± 0.008 µg Hg/g). Panel B of Figure 2 shows that addition of 100- or 300-mM Se solutions synergistically increased Hg contents of the leaves when Hg was also added. Supplementation with 100- or 300-mM Se resulted in 6- and 50-fold increases in Hg accumulation in leaves. Once again, until the roots are analyzed, it will not be known whether these results reflect an increased uptake from the soil or an increased redistribution of Hg from the roots.

Twenty-seven individual 4-inch-diameter *E. crassipes* plants were obtained from Pond Factory Plants. Individual plants were transferred to trace metal-free plastic containers prepared with 1500 mL of a 1:1500 solution of DynaGro. The plants were weighed and randomly assigned to Hg and Se treatment groups (three plants/group; 27 total plants) in a fully crossed 3 × 3 factorial (nine growth treatment groups) study of time- and dose-dependent interactive effects of these elements. Plants were supplemented weekly with 20 mL of either low, medium, or high Na_2SeO_4 (0.1, 1.0, and 10 mmol Se/kg, respectively) and 20 mL of low, medium, or high $HgCl_2$ (0.1, 1.0, 10 mmol Hg/kg, respectively). Plant growth was monitored during the ~8 week treatment study. At the end of the study, the entire plant was harvested and divided into leaves, stems, and roots that were stored frozen in trace metal-free plastic bags at -85°C until ready for analysis.

Total Hg and Se mass concentrations (parts per million) for each sample were converted to molar concentrations (micromole per kilogram). Means and standard deviations of molar concentrations of Hg and Se were calculated and graphed for each treatment group. Elemental concentrations of Hg and Se in tissues of different dietary treatment groups were analyzed using ANOVA to assess Hg–Se interactions and compared using t-tests. The amount of Se incorporated into leaves of *E. crassipes* grown in water that was treated with low levels of Hg and Se was 0.36 ± 0.06 µmol Se/kg (0.030 ± 0.004 µg Se/g). As seen in Panel A of Figure 3, when solutions containing an additional 100 or 300 mM Se were added, the amount of Se in the leaves increased 10- and 20-fold, respectively. Increasing Hg exposure resulted in increased Se distribution into the leaves when Se was low, but leaves of plants supplemented with 100-mM

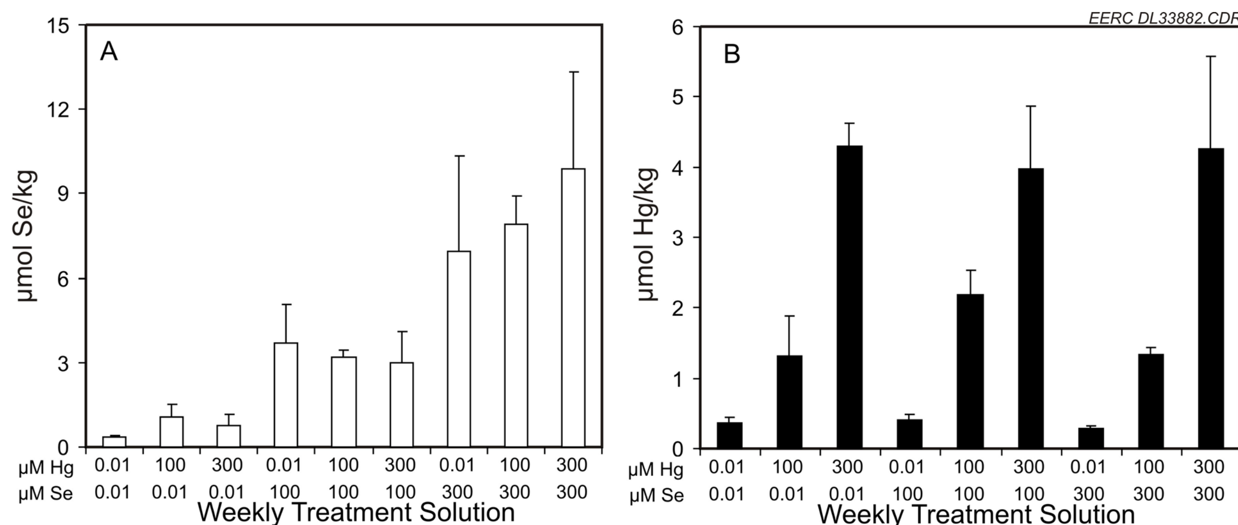

Figure 3. Mercury and selenium in E. crassipes grown in water treated weekly with solutions containing the indicated concentrations of Hg and Se.

Se solutions tended to have diminishing leaf Se in the presence of increasing Hg. Leaves of plants supplemented with 300 mM Se were observed to have more Se as increasing Hg was added. Further work will be needed to determine whether this indicates formation of HgSe complexes, as hypothesized, or whether these results reflect a shift in compartment partitioning that increased Se distribution into the leaves. Leaves of plants grown in water without added Hg or Se contained 0.37 ± 0.08 µmol g/kg (0.070 ± 0.02 µg Hg/g). Plant incorporation of Hg was less sensitive to Se-dependent effects than Se incorporation was to Hg effects (Panel B of Figure 2). Amounts of Hg incorporated appeared to be maximal in all groups independent of added Se, so increases in its rate of uptake may not have been possible to observe using the conditions employed in this study. The roots and stems from the plants in these treatment groups will be analyzed to determine whether similar effects occurred throughout the plant or varied by compartment.

MeHg–Se Interactions at the Cellular Level

The approach for this study was to use cell cultures grown in the presence of media containing graduated concentrations of Se and subject them to MeHg and inorganic Hg added at incremental log concentrations ranging from unexposed to toxic levels. Parallel studies of these Se-conditioned and Hg-exposed cultures are being challenged for specified time periods to establish the time- and dose-dependent effects of MeHg toxicity and Se's protective effects against toxicity. The end points measured include cell proliferation rates, viability, mercury, and selenium concentrations in the cells, Se-dependent enzyme activities, and rates of MeHg demethylation.

The cell line growth curve of human neuroblastoma SH-SY5Y cells administered specified concentrations of methylmercury, and methylmercury chloride has been established and is shown in Figure 4. SH-SY5Y cells were grown in Dulbecco's modified Eagle's medium–nutrient mixture F12 ham (DMEM-F12) supplemented with 10% fetal bovine serum. When the cells had reached 80% confluence, the cells were incubated with MeHg or mercury chloride for 24 hours. MeHg was administered at 0, 1, 5, 10, or 50 µM, and mercury chloride was administered at 0, 1, 5 and 10 mM. The viability of SH-SY5Y

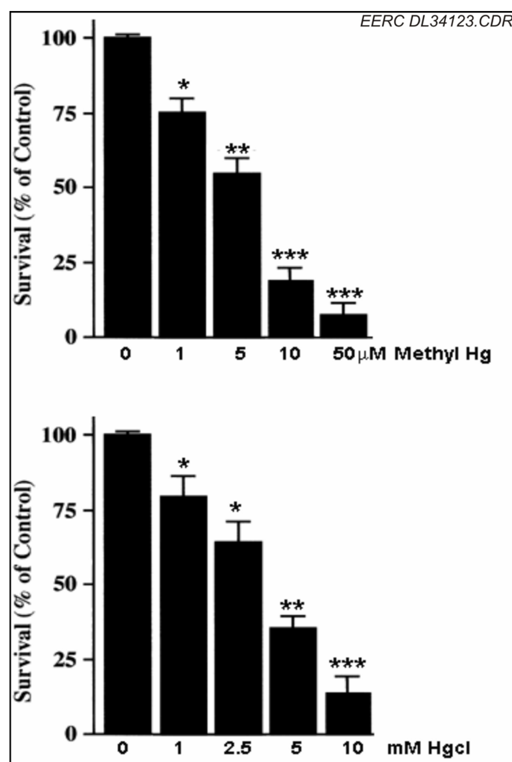

*Figure 4. Growth rates of human neuroblastoma SH-SY5Y cells administered the indicated concentrations of methylmercury and mercury chloride (*p<0.05, **p<0.01, ***p<0.001 [Student t test]).*

cells was determined by adding MTT to the SH-SY5Y cell cultures to reach a final concentration of 1 mg/mL according to the manufacturer's recommendations. Values obtained in the presence of MeHg or mercury chloride were normalized against values obtained in the absence of treatments. Three repeats were carried out for each measurement.

The approach was to use unsupplemented media as the basal concentration and then supplement with various concentrations of Se to determine the protective effects. The range of sodium selenite to be used in the studies has been established using prepared stock solutions of sodium selenite in PBS: Flask 1 – 0 added; Flask 2 – 0.5 µM; Flask 3 – 1 µM; Flask 4 – 2 µM; Flask 5 – 3 µM; Flask 6 – 5 µM; Flask 7 – 10 µM. Cell counts and viability were determined each day for 7 days.

Mercury's Effects on Cardiovascular Disease

Sample aliquots of ~0.5 g weighed to 0.0001 g from each heart sample were transferred into single-use, trace element-free 50-mL digestion tubes, with every tenth sample being prepared in duplicate and with elemental spike recovery samples being performed accompanying each batch. Each digestion batch included blank and certified standard reference materials (dogfish muscle certified reference material DORM-2).

Samples were prepared and analyzed using the following procedure:

1. Samples were treated with 5 mL of 16 N nitric acid (trace metal grade) and heated at 85°C in deep cell hot blocks fro 24 hours in capped tubes to preserve samples from trace element contamination.

2. Samples were cooled, and 1.5 mL of 30% hydrogen peroxide (Americal Chemical Society-certified) was added, and samples were recapped and returned to heating in the dry block at 85°C.

3. Samples were cooled, and 15 mL of 12 N HCl (trace metal grade) was added.

4. Samples were heated at 90°C for 90 minutes in order to reduce Se(VI) to Se(IV).

5. Samples were cooled and diluted to 50 mL with double distilled water.

6. Samples were analyzed for Hg content by cold-vapor atomic absorption spectrophotometry using a CETAC M-6000A, and Se was analyzed by hydride generation atomic fluorescence spectroscopy using a PS Analytical Dual Millennium Excalibur.

Mercury and selenium concentrations in normal hearts and hearts removed from patients with DCM or IHD are shown in Figure 5. Normal hearts contained 3.73 ± 0.54 µmol Se/kg and 0.47 ± 0.16 µmol Hg/kg (0.295 ± 0.43 µg Se/g, 0.093 ± 0.033 µg Hg/g). No age- or sex-related differences in tissue trace element concentrations were noted in normal or diseased patient groups. Hearts of patients with DCM contained 3.65 ± 0.59 µmol Se/kg and 0.39 ± 0.29 µmol Hg/kg, and hearts of patients with IHD contained 3.50 ± 0.43 µmol Se/kg and 0.17 ± 0.11 µmol Hg/kg.

No significant differences in selenium contents were noted between any of the study groups. Compared to normal hearts, mercury contents of DCM hearts were not significantly different, but hearts of patients with IHD contained significantly less mercury ($p<0.000001$). Mercury contents of hearts from IHD patients were also lower than hearts of patients with DCM ($p = 0.01$).

In contrast to the previous investigation (9) that noted extraordinarily high mercury contents in heart tissues of patients with DCM, this study finds the concentrations of mercury in DCM and normal hearts are indistinguishable. However, heart tissues from the IHD population used as a control group were found to contain significantly less mercury than was present in normal or DCM hearts. Since tissue mercury levels are usually directly related to seafood consumption, the low mercury levels noted in hearts from IHD patients may indicate low seafood consumption by this population.

It is unknown why the Frustaci investigation (9) observed mercury concentrations that are so much higher than this study, but the small sample mass (~1 mg) of the biopsy would greatly increase the effect of any contamination artifacts encountered during sample processing. Since the reported mercury concentrations of the heart biopsy tissue samples increased as ejection fraction decreased, it is also possible a component of the radio-opaque solution used for positioning biopsy sampling instruments may have contaminated the biopsy sample. These solutions may have either contained or appeared to have contained mercury. This possibility is being investigated.

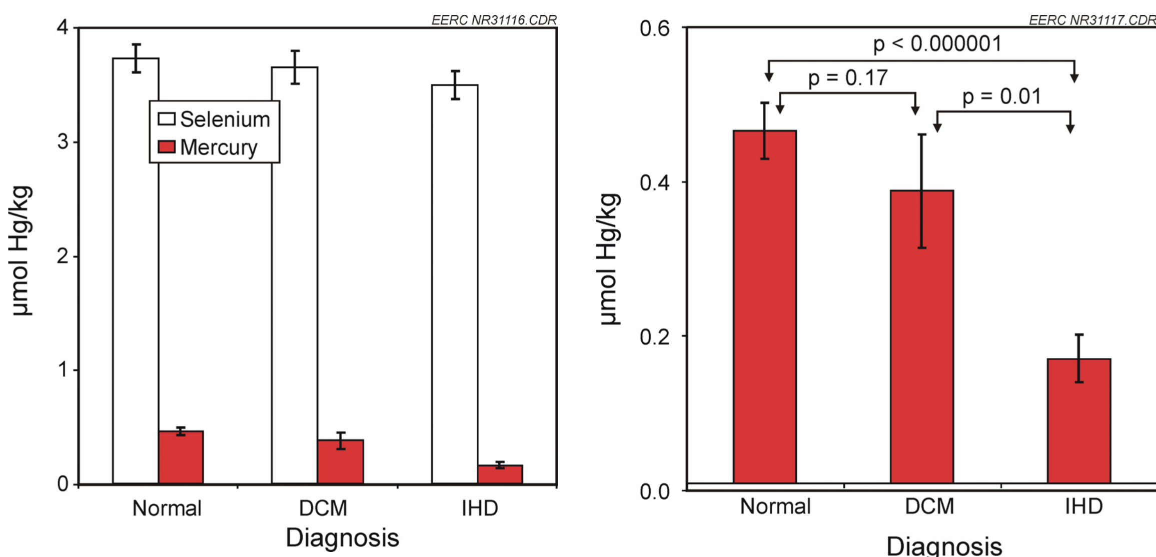

Figure 5. Mercury and selenium concentrations in normal hearts and hearts removed from patients with DCM or IHD.

Quality Assurance/Quality Control

An EERC quality management system (QMS), authorized and supported by EERC managers, is in effect and governs all programs within the organizations. Additionally, the CATM Program has a quality assurance plan (QAP) in effect that addresses trace metal emissions research at the EERC. The CATM QAP has been reviewed and accepted by EPA. This project follows the quality manual in order to obtain statistically valid and physiologically meaningful results regarding the interactions of mercury and selenium.

Quality Objectives

The quality objectives of all tasks are to conduct the studies with carefully controlled regimes so that they are repeatable; so that it is possible to clearly distinguish differences in the assessed end points; and to obtain analytically accurate and precise determinations of elemental contents.

Measurement/Data Acquisition

To validate consistency and validity in cell culture studies, the parameters are measured and the concentrations and time-dependent protective effects of selenium against Hg toxicity are evaluated and individually plotted. Multiple independent assessments are performed, and a minimum of three separate experimental collections of data points for each treatment group is included in determining the mean values and standard deviations for the survival and propagation effects. Selenoenzyme activity in cultured cells will be similarly measured in multiple independent assessments using established kit methods and their accompanying pool standards and controls, independent measurements of three or more sets of samples assessing total Hg, Se, and MeHg contents will be performed using large-scale batches of cultured cells to permit parallel assessments of total Hg and organic Hg. In all studies, trace elements are

determined in batches that include certified QC samples analyzed relative to calibration standards according to established protocol.

Assessment and Validation

Protocols being used in this project have been established and are maintained to ensure accurate and precise analytical results are obtained. All sampling, instrument calibrations, and QC considerations are included in the protocols. QC samples including analytical blanks and certified reference materials are included in each batch to ensure validity of observed analysis values.

Status

The project is currently ongoing. All tasks other than cell culture studies have been finalized, and the remaining cell studies are near completion. Final reporting and writing have begun, and material is being prepared for presentation at various conferences. Likewise, this information will be disseminated to support further related research studies.

Potential Applications and Benefits

Potential Users and Real-Life Applications

The findings of these studies will provide important information for EPA, the U.S. Department of Energy, the U.S. Food and Drug Administration, and the World Health Organization. This information may assist these agencies in making regulatory policy decisions regarding mercury exposure. Assessing selenium's involvement in mercury toxicity issues will help these agencies assess the risks of human mercury exposure and identify populations at risk and potential methods for protection and remediation.

Environmental and/or Health Benefits

The availability of selenium for environmental uptake may have an important effect on bioaccumulation of Hg in the food chain. Likewise, the formation of HgSe complexes within the biota may be involved in Hg phytoremediation. In ecosystems where the geological and biological availability of Se supports increased Se physiology, increased HgSe formation may contribute to increased Hg retirement. In ecosystems where Se either is absent from soils or is poorly available because of low pH or other limiting factors, the paucity of intracellular Se will limit Hg retirement as HgSe formation. These aspects may have implications for environmental treatments of high mercury remediation and bioaccumulation. The low mercury levels noted in hearts from IHD patients may indicate low seafood consumption by this population. This may have relevance in determining the benefits of increased fish consumption on cardiovascular health.

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